

# *Crocidura ramona* sp. nov. (Insectivora, Soricidae): a new species of shrew from the central Negev desert, Israel

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# Abstract

A new species of *Crocidura* (*C. ramona*) from Central Negev desert, Israel is described. The new species has small size, flattened skull, and light silver-gray dorsal pelage coloration. The diploid chromosome number is 28, NFa = 42, X-chromosome – acrocentric. The species belongs to the Palearctic group of the genus *Crocidura*. Within this large group, the nearest relative of *C. ramona* is *C. pergrisea*.

# Introduction

For decades, only three taxa of the genus *Crocidura* have been described in the fauna of Israel: *C. leucodon judaica* Thomas, 1919; *C. russula monacha* Thomas, 1906; and *C. suaveolens portali* Thomas, 1920 (HARRISON 1964; ATALLAH 1977; MENDELSSOHN and YOM-Tov 1987). The last taxonomic revision based on chromosomal, biochemical, and morphometric data (CATZEFLIS et al. 1985) presented evidence of conspecifity of *C. russula monacha* and *C. suaveolens*. This point of view was accepted, and now only two species of *Crocidura* (*C. leucodon* and *C. suaveolens*) are included in the list of Israeli mammals (HARRISON and BATES 1991; SHALMON 1993).

During the course of regular trapping sessions of rodents, lizards, and insects in the Central Negev, between 1993 and 1995, we found seven specimens of *Crocidura* clearly different from known species, both morphologically and karyologically. Investigation of zoological collections revealed four additional specimens of the same taxon. The new species, *Crocidura ramona* from the Negev Desert, is described.

# Material and methods

Two males, one female and one embryo (also female) of *Crocidura* sp. were used for karyotype analysis. The chromosomes of adult animals were prepared by the standard method from bone marrow and spleen cells. Chromosomes from embryos were obtained from liver tissue by use of a short-term cell culture method: the liver cells were incubated at 37 °C for two hours in Medium 199 and calf serum (Sigma) at a proportion of 6:1. Then two drops of 0.004% colchicine solution were added to 6 ml of the suspension, and after 20 minutes the conventional chromosomal technique was employed. Chromosome preparations were stained with 4% Giemsa. For G- and C- band staining, the trypsin (SEABRIGHT 1971) and BSG (SUMNER 1972) methods were applied. As comparative material we used G-banded chromosomes of one male of *C. suaveolens* captured on Mt. Carmel, Israel.

Skull morphology was studied on eight specimens of *Crocidura* sp. trapped in the Makhtesh Ramon and in the vicinity of Mizpe Ramon, Central Negev desert. These were then compared with 47 specimens of *Crocidura suaveolens* and 12 specimens of *Crocidura leucodon* from zoological collections at Tel-Aviv (TAU) and Hebrew (HU) Universities. Four cranial measurements were analysed: condylo-incisive length (Lci), zygomatic breadth (Bz), preorbital breadth (Bpo), and skull height (H). All measurements were performed according to KAHMANN and VESMANIS (1974) and ZAITSEV (1991).

## **Results and discussion**

#### Skull morphometry

Measurements of the examined specimens are presented in table 1. All of the *Crocidura* specimens from the vicinity of Mizpe Ramon (seven of ours and one from the HU collection) are clearly different from both *C. leucodon* and *C. suaveolens* in skull size and proportions. The specimens of the Central Negev are much smaller than *C. leucodon*, but similar in size (or slightly smaller on average) to *C. suaveolens*. Likewise, skulls of the specimens from the Central Negev are flattened; this character is shared with *C. leucodon*, but different from *C. suaveolens* (Fig. 1). Three additional specimens, indistinguishable from the Mizpe Ramon sample, were found in the TAU collections: two from Sede Boqer (Central Negev, 30 km north of Mizpe Ramon) and one from Sartaba (northern edge of Judean Desert).

**Table 1.** Skull measurements (upper line – min–max, lower line – mean ± SD) of the three *Crocidura* species of Israel. Skull measurements: condylo-incisive length (Lci), zygomatic breadth (Bz), preorbital breadth (Bpo), skull height (H), and relative skull height index (H/Bpo)

Species	Lci	Bz	Вро	Н	H/Bpo
C. suaveolens $(n = 47)$	17.0 - 20.0	5.2 - 6.3	3.0 - 3.6	3.1 - 3.8	0.97 – 1.12
	$18.69\pm0.62$	$5.79 \pm 0.23$	$3.33\pm0.14$	$3.46 \pm 0.15$	$1.041 \pm 0.035$
<i>C. ramona</i> sp. n. (n = 11)	17.7 –18.4	5.1 - 5.6	3.2 - 3.6	3.1 – 3.3	0.86 - 0.97
	$18.05 \pm 0.26$	$5.37 \pm 0.15$	$3.44 \pm 0.13$	$3.15 \pm 0.07$	$0.919\pm0.031$
C. leucodon $(n = 12)$	19.7 - 21.7	6.2 - 6.8	3.4 - 4.0	3.4 - 3.8	0.91 - 1.06
	$20.78\pm0.56$	$6.53\pm0.20$	$3.73\pm0.20$	$3.61\pm0.12$	$0.970 \pm 0.041$

## Karyotype analysis

All karyotyped specimens of *Crocidura* sp. have 28 chromosomes in the diploid complement (Fig. 2 a). Three groups of chromosomes can be distinguished based on their morphology: the first group consists of three pairs of submeta- subtelocentric chromosomes; the second group includes five pairs of subtelocentrics; and the third group involves five acrocentric pairs (NFa = 42). The X-chromosome is acrocentric, comparable in size with the first chromosome pair of the second group. The Y-chromosome is acrocentric, equal in size with the last acrocentric autosomes. The karyotype of *Crocidura suaveolens* from Mt. Carmel has 40 chromosomes: four pairs of subtelo- submetacentric autosomes, 15 pairs of acrocentric autosomes and submetacentric X-chromosome and acrocentric Ychromosome (NFa = 46). The karyotype of *C. suaveolens* from Mt. Carmel does not differ from the chromosome complements of *C. suaveolens* that have been studied from the vicinity of Tel-Aviv (CATZEFLIS et al. 1985) and from other localities (CATZEFLIS et al. 1985; REUMER and MEYLAN 1986; IVANITSKAYA 1989; GRAFODATSKY et al. 1991).

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Fig. 1. Scatter-plot of cranial measurements of the three Crocidura species of Israel. Lci – condylo-incisive length, H/Bpo – index of relative skull height (skull height/preorbital breadth). Solid circles – C. ramona sp. nov., open circles – C. suaveolens, triangles – C. leucodon. Dotted ellipses confine the 95% confidence areas.

The C-band method revealed a very small amount of autosomal heterochromatin in *Crocidura* sp., localized in the pericentromere regions of 9 chromosome pairs with different degrees of expression. The short arms of the first pair of subtelocentric autosomes are almost entirely C-positive. The X-chromosome does not contain the heterochromatin material, and the Y-chromosome completely consists of heterochromatin (Fig. 2 b). All species of *Crocidura* studied with the C-banding method have a small amount of heterochromatin in the autosome complement and C-positive Y-chromosomes (HARADA et al. 1985; TADA and OBARA 1986; GRAFODATSKY et al. 1988; IVANITSKAYA 1989).

At the present time the karyotypes of 50 species of the genus Crocidura are known. Most of these species are differentiated karyologically. Intraspecific chromosomal variability has been revealed in six species (MADDALENA and RUEDI 1994; RUEDI and VOGEL 1995). C. suaveolens is the widest geographically distributed species of the genus; of more than 30 karyologically studied populations of this species, only four possess chromosomal variability. Three populations from Switzerland are polymorphic for the number of chromosomes, due to the presence in some karyotypes of B-chromosomes (MEYLAN and HAUSser 1974). In addition, one population from Japan is characterized by 2n = 39 - 40, as a result of a Robertsonian translocation (TSUCHIYA 1987). The other chromosomally polymorphic populations of Crocidura occur only in Southeastern Asia and in the Afrotropical region. The diploid numbers in *Crocidura* species vary from 2n = 22 in *C. pergrisea* to 2n = 60 in C. cf. bicolor and NF - from 34 to 86. The high numbers of 2n and NF are typical for species of Afrotropical origin; species with low chromosomal numbers are distributed in the Palearctic and Oriental regions (MADDALENA and RUEDI 1994). Thus, Crocidura sp. belongs to the second group by virtue of its chromosomal formula. C. leucodon also has the same number of chromosomes (2n = 28) as Crocidura sp., but

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Fig. 2. Conventionally stained (a) and C-banded (b) chromosomes of *C. ramona* sp. nov. (male, no. M-9342 (TAU), holotype).

these two species vary distinctly in their number of chromosomal arms: NFa = 42 in *C. ramona* and NFa = 52 in *C. leucodon*. In addition, *C. leucodon*, like most *Crocidura* species, has a biarmed X-chromosome. To date, an acrocentric X-chromosome has been revealed only in two species: one Palearctic, *C. pergrisea* (GRAFODATSKY et al. 1988) and one Oriental, *C. lepidura* (RUEDI and VOGEL 1995).

Comparative analysis of G-banded chromosomes of *Crocidura* sp. and *C. suaveolens* revealed similar G-band patterns in some chromosomes (Fig. 3). All acrocentric chromosomes, the Y-chromosome and two pairs (1 and 3) of the first group of *Crocidura* sp. have a banding pattern that is practically identical to the corresponding chromosomes of *C. suaveolens*. Subtelocentric chromosomes of *Crocidura* sp. match only partially with *C. suaveolens* chromosomes. The acrocentric X-chromosome of *Crocidura* sp. has inverted G-band patterns in comparison to the biarmed X-chromosomes of *C. suaveolens*. The same type of rearrangement in X-chromosome was recorded for *C. pergrisea* (GRAFODATS-KY et al. 1988). Apparently, the X-chromosomes of *Crocidura* sp. and *C. pergrisea* are closely related.



Fig. 3. Comparison of G-banded chromosomes haploid sets of *C. ramona* sp. nov. and *C. suaveolens*. In each pair the left chromosome represents *C. ramona* and the right one is of *C. suaveolens*. Unpaired chromosomes in the lower row represent chromosomes of *C. suaveolens* that have no analogs in the karyotype of *C. ramona*. Triangles mark the centromere positions.

It is not possible to derive the karyotype of *Crocidura* sp. directly from a karyotype of any recent species. Comparative analysis of G-banded chromosomes allowed MADDALENA and RUEDI (1994) to reconstruct the ancestral *Crocidura* karyotype with 2n = 38 and NF = 54 - 58. The chromosomal complement of *C. suaveolens* is close to this karyotype. The origin of *C. leucodon* (2n = 28) and *C. pergrisea* (2n = 22) karyotypes can be explained mainly by Robertsonian type of rearrangements (GRAFODATSKY et al. 1988). Five pericentric inversions (or centromere shifts) and five centromere-telomere fusions are necessary for evolving from the hypothetical ancestral karyotype to the karyotype of *Crocidura* sp. If we accept the inverted structure of X-chromosomes as an apomorphous character, *C. pergrisea* and *Crocidura* sp. can be considered as sister taxa within the Palearctic group of *Crocidura*.

Thus, both morphological and karyological analyses lead to the conclusion that *Crocidura* sp. represents a new species.

## Crocidura ramona, sp. nov.

**Holotype:** 3 adult, No. M-9342 deposited in TAU collection, NE region of Makhtesh Ramon, Negev (30° 40' N 34° 56' E), 1. 04. 1995, skull and skin.

**Paratypes:** ♂ adult, No. M-9343, TAU collection, central region of Makhtesh Ramon, Negev (30° 35' N 34° 51' E), 13. 03. 1993, skull; ♀, adult, No. M-9344, TAU collection, SW

region of Makhtesh Ramon, Negev (30° 30' N 34° 40' E), 4. 08. 1994, skull;  $\bigcirc$  adult, No. M-9345, TAU collection, northern rim of Makhtesh Ramon, Negev (30° 35' N 34° 42' E), 21. 01. 1995, skull;  $\bigcirc$  adult, No. M-9346, TAU collection, NE region of Makhtesh Ramon, Negev (30° 40' N 34° 56' E), 1. 04. 1995, skull, skin;  $\bigcirc$  adult, No. M-9347, TAU collection, central region of Makhtesh Ramon, Negev (30° 35' N 34° 50' E), 27. 04. 1995, skull, skin;  $\bigcirc$  adult, No. M-9348, TAU collection, central region of Makhtesh Ramon, Negev (30° 34' N 34° 50' E), 30. 04. 1995, skull, skin:

Additional material: No. M-3347 HU collection, Mizpe Ramon; No. M-8675, M-8676 TAU collection, Sede Boqer; No. M-8703 TAU collection, Sartaba.

**Diagnosis:** Shrews of relatively small size: condylo-incisive length of skull 17.7–18.4 mm. Skull is flattened: the ratio of skull height to preorbital breadth less than 1.00. Third upper molar is moderately reduced. Dorsal pelage coloration is light silver-gray. Chromosome set: 2n = 28, NFa = 42, X-chromosome: acrocentric.





Fig. 4. Skulls of C. ramona sp. n. (a) and C. suaveolens (b) in lateral view.

**Description and comparison:** Size is relatively small: external measurements (based on four specimens) are as follows: head and body length: 58–63 mm; tail length: 42–43 mm; hindfoot length: 10–11 mm; and ear length: 7.5–8.0 mm; body weight: 4.0–5.0 g. In comparison with other North African, Arabian, and eastern Mediterranean species (SPITZENBERGER 1971; OSBORN and HELMY 1980; HUTTERER and HARRISON 1988; HARRISON and BATES 1991; ZAITSEV 1991; HUTTERER 1992, 1994), *C. ramona* is much smaller than *C. leucodon*, *C. lasia*, *C. pergrisea*, *C. somalica*, and *C. olivieri*. Body length of *C. ramona* is smaller on average than *C. suaveolens* and *C. floweri*, equal to that of *C. religiosa*, but greater than *C. arabica*; however, it is still within the limits of variation for the first two species. Tail length of *C. ramona* is within the limits of variation for *C. suaveolens*, shorter than *C. floweri* and longer than *C. arabica* and *C. religiosa*. Hindfoot length of *C. ramona* is less than *C. suaveolens* and *C. floweri* and equal to *C. arabica* and *C. religiosa*.

The pelage coloration is light. The hairs are bicoloured throughout with slaty gray bases. The tips are silvery white on the ventral surface and silvery gray on the dorsal surface. The line of demarcation along the flanks is clear, but not very sharp. The tail is indis-



Fig. 5. Rostral part of skulls of C. ramona sp. n. (a) and C. suaveolens (b) in ventral view.



tinctly bicoloured, with its dorsal surface lighter than its back. In coloration, *C. ramona* is distinct from most *Crocidura* species, with the exception of *C. pergrisea*.

The skull (Fig. 4) is smaller than that of *C. leucodon*, *C. lasia*, *C. somalia*, and *C. olivieri*, it is larger than that of *C. religiosa*, and is approximately similar in size to *C. suaveolens*, *C. pergrisea*, *C. arabica*, and *C. floweri*. The skull is markedly flattened in

dorsal profile. The extent of this flatness in *C. ramona* is much greater than in most other *Crocidura* species, including *C. leucodon*, but less than in *C. pergrisea*.

The upper incisor  $(i^1)$  is powerful, as that of *C. leucodon*, and noticeably heavier than in other species. In the maxillary dentition, unicuspid<sup>1</sup> is the largest; the crown area of unicuspid<sup>2</sup> is smaller than that of unicuspid<sup>3</sup>, the character shared with *C. leucodon*, but not with other species. The internal cusp (hypoconus) of the upper premolar is very small, noticeably smaller than that of *C. suaveolens*, *C. pergrisea*, and *C. arabica*, but not completely reduced as in *C. leucodon*. The third upper molar is moderately reduced posteriorly (Fig. 5). It is smaller than that of *C. suaveolens* and *C. pergrisea*, but larger than that of *C. leucodon*, *C. arabica*, and African species.

**Etymology:** Named for the Ramon erosion cirque (Hebrew name – Makhtesh Ramon), Central Negev, Israel, where the new species was found.

**Distribution:** The species has been found in three regions: the vicinities of Mizpe Ramon, Sede Boqer, and Sartaba (Fig. 6). The first two regions are located on the Negev Highlands, and the third is on the north edge of the Judean Desert. All three regions are located in rocky desert, from 400 to 950 m a. s. l. We did not find any other *Crocidura* species co-occurring with *C. ramona*. All localities of *C. suaveolens* in Israel are confined to regions with a Mediterranean climate or to lowland deserts (i. e., Mediterranean sandy coastal plain, Rift Valley). The only shrew species found together with *C. ramona* was *Suncus etruscus*.

**Habitats:** Of seven specimens from the vicinity of Mizpe Ramon, five were caught in the dry riverbeds with *Retama* and *Tamarix* vegetation among gravel plain with rock outcrops. The last two specimens were caught near dry riverbeds with dense *Atriplex* vegetation among rocky hills, partially covered with loess.

**Remarks:** Based on its morphological and karyological characters, *C. ramona* undoubtedly belongs to the Palaearctic, but not to the Afrotropical, group of the genus *Crocidura* (in the sense of MADDALENA and RUEDI 1994). Within this large group, it is more or less equally distant from *C. suaveolens*, *C. leucodon*, and *C. pergrisea*. However, based on coloration, some skull characters, the structure of X-chromosome, and habitat distribution, it is possible to suggest that the nearest (but not closely related) relative of *C. ramona* sp. nov. is *C. pergrisea*. This being true, an ancestor of *C. ramona* penetrated into the territory of Israel from Asia Minor. This penetration presumably occurred in the Early Pleistocene, as the first records of *Crocidura* from Jordan Valley are dated to that time (TCHERNOV 1988).

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## Zusammenfassung

## Crocidura ramona sp. nov. (Insectivora, Soricidae): eine neue Spitzmausart aus dem Zentralteil der Negev Wüste, Israel.

Eine neue Art von *Crocidura* aus dem Zentralteil der Negev Wüste in Israel wird beschrieben. Diese neue Art ist durch kleine Körpergröße, flachen Schädel und silbergraue Färbung des Rückenfells gekennzeichnet. Die Art gehört zu der Palaearktischen Gruppe der Gattung *Crocidura*. Nächster Verwandter von *C. ramona* innerhalb dieser großen Gruppe ist *C. pergrisea*.

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